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BIOCHEMICAL STUDIES OF THE INFLUENCE OF DIET ON THE COMPOSITION AND BIOLOGICAL PROPERTIES OF EGGS

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THE AMINO ACID CONTENT OF EGGS AND CHICKS: RELATION TO DIET AND TO INCIDENCE OF CHONDRODYSTROPHY[1]

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Pollard and Carr ('24) and Gerber and Carr ('30) reported differences in the Van Slyke nitrogen distribution of eggs from pigeons on different diets. McFarlane, Fulmer and Jukes ('30) were unable to find any significant influence of diet on the amino acid content of hen eggs. Titus, Byerly, and Ellis ('33) reported evidence of a slight effect of the diet on the total nitrogen of the egg yolk. Calvery and Titus ('34) were unable to find any marked difference in the composition of the proteins of eggs from pullets on different protein diets.

Titus, Byerly and Ellis also found that certain protein diets tend to lower the hatchability of the eggs produced. Part of the chick mortality was shown to be caused by chondrodystrophy (Chondrodystrophia foetalis), a type of defective bone and cartilage formation of unknown etiology. Their evidence indicated an amino acid deficiency as a possible factor in the etiology of chondrodystrophy. Since this abnormality represents primarily a failure in cartilage development, we observed that gelatin, the best known protein in cartilage, contains about 22 per cent glycine. Corn, on the other hand, contains no glycine in its chief protein, zein.

[1] Paper no. 1382, journal series, Minnesota Agricultural Experiment Station. The data in this paper were submitted by A. Rae Patton to the Graduate School of the University of Minnesota in partial fulfillment of the requirements for the degree of doctor of philosophy, June, 1935. All pertinent individual data are available for reference from the Division of Agricultural Biochemistry.

A corn ration was found by Byerly, Titus and Ellis ('33 a, '33 b) to permit an exceedingly high incidence of chondrodystrophy. It is for this reason that the glycine content of eggs was investigated with reference to the incidence of chondrodystrophy.

EXPERIMENTAL

The following rations were used:

Optimum ration	*Per cent*
Ground yellow corn	20
Standard wheat middlings	20
Ground oats	20
Bran	10
Meat and bone meal	5
Fish meal	5
Soybean meal	5
Milk	5
Alfalfa leaf meal	6
Wheat germ	3
Cod liver oil	1
NaCl, $CaCO_3$, corn, wheat	ad. lib.

Deficient ration	*Per cent*
Ground yellow corn	70
Corn gluten (lot 39)	22
Cod liver oil	1
Wheat germ	3
Oat hulls	1.5
Monosodium phosphate	0.9
Calcium carbonate	1
Sodium chloride	0.5
NaCl, $CaCO_3$, corn	ad. lib.

Fourteen white Leghorn pullets were raised on the 'optimum' ration, and then restricted to the 'deficient' ration from May 4 to August 12, 1933. During this period average egg production dropped from eight eggs per day to less than two eggs per day. Eggs produced on the 'optimum' ration, and others produced near the close of the period on the 'deficient' ration were analyzed. The entire egg contents were extracted 48 hours each with acetone, alcohol, and ether, yielding samples containing 16.0 per cent nitrogen. In the case of chick samples,[2] the entire egg contents were treated in the same manner. A sample for analysis consisted of the contents of six eggs.

Tryptophane and tyrosine were determined by the method described by Folin and Marenzi ('29), histidine by the Rosedale-Da Silva method ('32), total surfur by the Denis method

[2] Chondrodystrophic material was identified and provided by Dr. F. B. Hutt, who also supplied the normal chicks for comparison.

('10). The results are given in table 1. Van Slyke nitrogen distribution was determined according to the Cavett modification ('32). The duplicate analyses are given in table 2. The glycine analyses shown in table 3 were made by the method described by Patton ('35), and the total creatinine by the method of Folin ('14). The glycine data for the egg

TABLE 1

Tryptophane, tyrosine, histidine and total sulfur determinations on eggs from hens fed optimum and deficient (corn) rations

	TRYPTOPHANE	TYROSINE	HISTIDINE	SULFUR
	per cent	*per cent*	*per cent*	*per cent*
Eggs from White Leghorns optimum ration	1.08 ± 0.06	4.27	2.26	1.3
Eggs from same flock after 14 weeks on corn ration	1.06 ± 0.05	4.28	2.35	1.4

TABLE 2

Van Slyke nitrogen distribution. The figures are expressed as percentage of total nitrogen

	EGGS FROM HENS FED				NORMAL CHICKS		CHONDRODYSTROPHIC CHICKS	
	Optimum ration		Deficient ration					
Ammonia	14.62	14.75	10.06	12.50	13.50	13.70	16.60	16.20
Humin	10.00	7.38	8.07	4.75	8.63	8.77	9.46	8.12
Filtrate amino	37.80	38.10	43.30	46.20	39.20	38.40	35.70	36.60
Filtrate non-amino	9.70	10.90	10.90	10.80	13.80	14.10	14.00	14.20
Arginine	9.75	9.75	11.00	11.00	8.77	8.77	8.77	7.80
Cystine	0.39	0.39	0.42	0.33	0.36	0.36	0.41	0.42
Histidine	6.40	6.40	3.90	4.55	6.90	7.60	5.40	6.70
Lysine	9.76	9.76	11.97	11.42	11.37	10.67	9.73	9.38
Total	98.42	97.43	99.62	101.55	102.53	102.37	100.07	99.42

samples represent the average of concordant results from single samples and for the chicks the mean of average data from two samples.

In addition to the samples described, eggs were collected from two white Leghorn pullets fed the optimum ration plus 1 gm. of glycine per day, in capsules. Glycine was determined in the eggs before and after 1 month of glycine feeding.

Glycine exerted toxic action in large doses (4 gm. a day). Two days of this treatment resulted in cessation of egg production. Food and water consumption were practically zero. Both hens exhibited extreme prostration, inability to stand, and sudden fits of agitation. When glycine feeding was discontinued, the symptoms rapidly disappeared, and normal egg production was resumed.

TABLE 3

Glycine and total creatinine determinations

	MEAN GLYCINE CONTENT OF TOTAL 'PROTEIN'	TOTAL CREATININE IN DRY MATTER
	per cent	*per cent*
Eggs from White Leghorns, optimum ration	2.21	
Eggs from same flock after 14 weeks on corn ration (glycine deficient)	2.16	
Eggs from hen R on optimum ration before feeding glycine	1.80	
Eggs from hen R on optimum ration plus glycine	1.87	
Eggs from hen L on optimum ration plus glycine	1.82	
Chick embryos which developed normally	9.5 9.1	0.02
Chick embryos which died from chondrodystrophy	6.8 6.3	0.02

DISCUSSION

On the basis of the glycine determinations certain conclusions may be drawn. In the first place, feeding a low glycine ration does not alter the glycine content of the eggs which the hen lays. Also, feeding a high glycine ration does not alter the glycine content of the eggs, either free or combined in protein. Since incubated eggs contain more glycine than fresh eggs, there appears to be a synthesis of glycine during embryonic development.

It is interesting that the total creatinine values found were the same in both normal and chondrodystrophic chick samples. This may constitute evidence of the similarity in age of the

chick groups compared, since total creatinine is known to increase rapidly during incubation (Needham, '31).

Any comparisons between the composition of the rations and of the chicks shown in table 2 would be purely theoretical, since the chondrodystrophic chicks were not produced on the 'deficient' ration used. The ammonia nitrogen values diverge in an unexpected fashion, as do the humin, filtrate amino, and lysine nitrogen values. The significance of this cannot be interpreted at the present time. Arrangements have been made for continuing the phase of this work concerning amino acids and chondrodystrophy.

SUMMARY

1. A significant difference was found between the glycine contents of normal and chondrodystrophic embryos.

2. A synthesis of glycine during development of the hen's egg was demonstrated.

3. Glycine was found to be toxic to hens when fed in large doses.

4. Feeding glycine to hens did not influence the glycine content of the eggs.

5. An investigation has been made of the amino acid content of eggs from fowls receiving 'optimum' and 'deficient' protein rations.

6. The analyses, including tryptophane, tyrosine, histidine, glycine, arginine, cystine, lysine and the Van Slyke nitrogen distribution, did not demonstrate a significant effect of the diet on the composition of the egg proteins.

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